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NOTES ON THE PARASITES OF THE LAKE FISH

III. ON THE STRUCTURE OF THE COPULATORY ORGANS IN *Microphallus* NOV. GEN.

By HENRY B. WARD.

WITH ONE PLATE

In a previous paper (94) published in the journal of this Society, I gave a brief account of the life history and structure of a new species of distome from the dogfish (*Amia calva* L.). Since that time our knowledge of the group of distomes has been extended greatly and it is possible to give a clearer idea of the position and relationship of this form. The papers of Looss, notably one of the most recent (Looss, 99) have contributed most prominently towards this end and have laid a firm foundation for a rational system among the distomes. It is also true that some of the references to this particular species have misinterpreted in certain points the brief description given previously and have thus rendered it more important to rediscuss its structure and affinities. At the same time the type of structure presented by the copulatory organs, which Looss has shown to be of prime importance in the taxonomy of the group, differs in an interesting manner from any plan for these organs given by that author and forms a possible transition towards the type described by Jägerskiöld (00) for another genus.

Besides the original description of the species (Ward, 94) the following references have been made to it: MacCallum (95) merely records its presence in the stomach of *Anguilla chrysopa*, a new host. Stossich (99) in his revision of the genus *Brachycoelium* Duj. founded a new genus, *Levinsenia*, and included in it the four species, *L. opacum* (Ward 1894), *L. brachysomum* (Creplin 1878), *L. pygmaeum* (Levinsen 1881) and *L. macrophallos* (v. Linstow 1875). He did not, however, specify

the type form of this new genus, so that Luehe (99), while questioning the formation of the genus with the limits given by Stossich, deemed it nevertheless advisable to designate a type species in order to avoid so far as possible the confusion incident upon later revision of the group, and he therefore (note 30, p. 538) proposed *D. brachysomum* as type.

Somewhat later Jägerskiöld (00) gave a careful description of the structure of *L. pygmaea* together with a revised diagnosis of the genus, basing the same on *L. pygmaea* which is specified as type. It is unfortunate in view of the accurate and full knowledge we possess regarding this species that its designation as type comes too late to be accepted. Stiles* has recently called attention to the fact that the name *Levinsenia* is preoccupied and has renamed the genus under discussion *Levinseniella*.

Looss (99) in his discussion of the paper by Stossich (99) criticises the inclusion of *D. opacum* in the genus *Levinsenia*, and after comparing its structure with that of the other species grouped by Stossich within this genus concludes the discussion with the following words: "Unter solchen Umständen glaube ich das *Dist. opacum* Ward als Typus einer eigenen Gattung betrachten zu sollen, die nahe Verwandtschaft zu *Lecithodendrium* zeigt, wohingegen das Genus *Levinsenia* Stossich auf die Formen vom Typus des *D. brachysomum* Crepl. zu beschränken wäre" (p. 621). In the following pages I hope to show that the precision which Looss has manifested in other cases in evaluating generic relationships has been equally clearly exercised here, and that further knowledge of the structure of this species fully justifies its position in an independent genus, for which I propose the name of *Microphallus*.

For general features in the structure of this species reference should be made to the earlier paper (Ward, 94) and to the figure of the adult which is reproduced here (Pl. XXVI, Fig. 1). With regard to the terminal region of the reproductive system some additional facts of importance are to be discussed here. The common sexual pore lies at the left of the acetabulum just behind the middle of that organ, but separated from it by an appreciable distance. Furthermore, as a cross section shows distinctly, both the genital sinus and the ducts are independent of the ventral sucker and the ducts rise

* Notes on Parasites Nos. 56-58. In MS., to be published soon in "Science."

to the surface from the deeper lying organs in a direct line rather than as in *Gymnophallus* (Odhner, 00) following the outline of the sucker. The genital sinus is small, with hardly more than space for the copulatory organ to be described later, and its margin is so little distinct that one often finds in section a rounded depression without any projecting lip at all. The ductus ejaculatorius lies nearer the acetabulum with the orifice of the flattened metraterm on its outer (lateral) face at the base of the copulatory papilla. While in section the metraterm appears as a flattened duct with its walls in contact and with at most a meager slit-like lumen in spite of the comparatively well developed muscles of the wall, the adjacent ductus ejaculatorius manifests ordinarily a gaping circular lumen. This is particularly true in the heavy-walled copulatory papilla which terminates the duct. There are no special features in the structure of the metraterm or of other parts of the female reproductive system which has been sufficiently described in the previous paper. The male organs, however, display some features, the structure and significance of which were not understood before.

The form of the seminal vesicle is unusual, in that it does not consist of an enlarged canal thrown into two or more loops as in most distomes, but is rather of a nearly spherical form. In all individuals examined, save those actually *in copula*, it was filled so full of spermatozoa that one could see the structure of the wall only in occasional regions. I was unable to identify muscles of any sort, but could recognize occasional nuclei in the otherwise unbroken membrane. In those individuals which were copulating the walls of the vesicle appear collapsed and irregular in outline and the mass of included sperm was much reduced in size. It is, however, difficult to reconcile the appearance of the walls of the vesicle with muscular or elastic contraction on their part; one is impressed with the idea that they have been passively collapsed by the action of the body muscles, especially the oblique, which appear tense, while the walls of the vesicle certainly look as if they had crumpled under external pressure.

The orifice of the vasa deferentia which join just as they reach the vesicle is so small and inconspicuous that I have as yet been unable to identify its character and cannot affirm or deny the presence of the valve cells described by Looss as found at this point in other species; certainly they are small

if present and must close the orifice very perfectly. On the other hand, similar cells are plain at the origin of the ductus ejaculatorius which starts from the vesicle at the posterior ventrolateral margin and passes almost directly ventrad to the copulatory papilla. The duct is short, measuring from 0.1 to 0.13 mm. in length, and has a delicate wall in which one can faintly see in favorable sections the cut ends of a row of delicate circular muscles; longitudinal fibres I could not detect. In spite of its shortness two regions can often be distinguished; that nearer the vesicle has a slight bulbous expansion and may properly be designated as the pars prostatica. The gland cells themselves appear in section as a crescentic mass around the origin of the duct from the vesicle (Fig. 3) and as they manifest a predilection for haematoxylin stains are a prominent feature in sections stained by such fluids. It should be noted, however, that in proportion to the ordinary development of these glands, the size of the organ in this species is actually insignificant and it is visible only on the most successful toto preparations.

When the duct reaches the base of the copulatory papilla, the lumen is slightly contracted and then suddenly expands into a flask-shaped cavity in the papilla, at the apex of which it opens into the genital sinus. It is this copulatory papilla which is the most peculiar structure in this form. When fully retracted it presents the appearance of a conical pyramid (Figs. 3, 4) somewhat longer than broad and irregular in outline. Its length varies from 50 to 60 μ and its transverse diameter at the point of greatest thickness is not more than 40 μ . When emitted (Fig. 2) the form is decidedly more elongate in appearance, though its actual length is approximately the same (55 μ). The lumen, however, is narrower at the proximal portion of the organ and broader near the tip, while the walls are but half so thick. The conical form of the retracted organ has also been modified to a tubular one, in which, however, a broader proximal and a narrow distal portion can be distinguished. The wall of the papilla is lined inside and out by a sharply defined clear membrane, the nature of which could not be further determined. The substance of the wall is composed of thickly set transverse fibres, among which neither nuclei nor other structures could be distinguished. In general appearance and reaction towards staining fluids these fibres resemble muscles, but their direction and arrange-

ment are not easily explained on such a hypothesis. There is present also a set of delicate muscular fibres surrounding the cloaca; they originate around the base of the papilla, from which they diverge at varying angles toward the points of their insertion into the skin. These fibres can be identified positively only in the most favorable cases, on account of their delicacy and sparsity. Their function in the emission of the organ is evident and when powerfully contracted thereby they appear very plain (Fig. 2). The invariable presence of this structure, which I have called a copulatory papilla, precludes the possibility of regarding it as an evertible cirrus which by chance was protruded in the specimen described.

Looss has called attention (94, p. 196) to the importance of determining the changes in form and position of the various parts of the reproductive ducts during copulation. A number of pairs taken *in copula* and preserved at once, enabled me to determine very precisely these relations for this species. It should not be forgotten that the peculiar character of the copulatory organs here make it impossible to draw any conclusions regarding other forms. When paired the worms are united by the ventral suckers so firmly that they cannot be forced apart without tearing the tissue of the body. In each sucker lies a knob of the body wall of the other worm and the powerful contraction of the circular muscles at the margin of the sucker is shown by their prominence in the sections (Fig. 2). The genital sinuses are reduced in depth and their margins are nearly in contact; there are, however, no muscles or other structures to make these margins definite, and I am inclined to think that they play a purely incidental part in the process. The two copulatory papillae lie side by side, their adjacent walls in close contact and the tip of each inserted in the orifice of the metraterm of the other individual. Both the duct and the metraterm are expanded to the fullest extent and a continuous stream of sperm is passing from the vesicle of each into the uterus of the other individual. Reference has already been made to the crumpled appearance of the wall of the vesicle and the probability that it is contracted by extraneous rather than by proper muscles.

The description of the structure of the copulatory papilla will make it clear that the suggestion of Looss in regard to its morphological value can not be accepted. He says (99, p. 621) "Das, was Ward bei Abwesenheit eines echten Cirrus-

beutels als Copulations-organ, als 'morphologisches Aequivalent des Cirrusbeutels'* beschreibt und abbildet, duerfte kaum etwas anderes sein als der etwas vorgestuelpte Boden des Genitalsinus; moeglich, dass hier aehnliche Verhaeltnisse vorliegen wie bei *Lecith. sphaerula*." The description already given will evidently not admit of interpreting the organ as merely an evaginated portion of the genital sinus, and its invariable presence and uniform character forbid one to regard it as a temporarily everted structure. It is a constant organ and yet of an unusual type, as is seen from a comparison of the synopsis of conditions found in the male organs, given by Looss (99, p. 551). Since there is no trace of a muscular cirrus sac or of a connective tissue envelopement about the terminal organs, one is compelled to assign this to the first type, as given by Looss, in which, however, he states that the ductus ejaculatorius has the form of a simple tube without evertible portions. No mention is made of the presence of a terminal papilla, and I have also examined with great care specimens of *Opisthorchis** which Looss assigns to this type and find no trace of any structure in the region in which this papilla is found; the duct terminates without any modification whatever.

The search for a similar structure in some other distome has been conducted with difficulty. So far as the older literature is concerned, most figures and descriptions are silent or lacking in respect to the precise form of the terminal organs of the reproductive system. Even where in a work of later date and greater accuracy a projecting papilla is drawn at the end of the ductus ejaculatorius, e. g. in *Monostomum lacteum* (Jägerskiöld, 96, pl. IX, fig. 7) it is impossible to judge whether the structure is temporary or permanent, and if the latter, how far it corresponds to that under discussion. The case just cited occurs moreover among forms so distantly related that the two could be considered at most as convergent structures.

In only one of the true distomes has anything similar been

* My statement was really "of the cirrus," not "of the cirrus sac." Cf. also p. 182.

† The form examined was that described previously (Ward, 95) as *Distoma felineum* Riv. Further study convinced me that the differences noted in the paper between it and the true *D. felineum* were too great to be varietal, and Looss (99, p. 675) is right in stating that on the basis of the vitellaria alone the form should be regarded as an independent species. I take this opportunity of proposing for it the name of *pseudofelineum*. The description contained in the paper cited will suffice for its identification. In its proper genus it becomes *Opisthorchis pseudofelineus*.

described so far as I can ascertain, and even here the likeness is not more than a general one. It is in *Levinseniella* (*Levinsenia*) that Jägerskiöld (00) has described a conical body of muscular character which projects into the genital sinus and through which the terminal portion of the duct runs in an oblique manner, opening on the lateral face of the cone at about mid-height (Fig. 5). It is possible that the contraction and folding of an elongated organ might produce the conical form with lateral orifice which this author has described in *Levinseniella*, and the prominently striated appearance he has represented recalls the structure described above. But even at best there is a striking difference in the form as well as in degree of development manifested by the organ in the two species. In *Levinseniella* it is equal in size to the ovary and to the ventral sucker and is a prominent feature in the general external appearance of the animal. In *Microphallus* on the other hand it is so small as to be seen only in sections. A comparison of the figures given by Jägerskiöld and myself will make this difference apparent at once. The discussion of the further differences between the two genera is reserved for a later paragraph. Although genuine differences exist between the conical body of *Levinseniella* and the copulatory papilla of *Microphallus*, I share fully the opinion expressed by Jägerskiöld that the latter is a low development stage of the former, and believe that the description of the structure given above raises this view from the rank of conjecture to that of acceptable homology.

In another respect this organ furnishes a decided addition to our knowledge of the distomes. In demonstrating the presence of a permanent muscular organ projecting into the genital sinus and carrying the terminus of the male duct Jägerskiöld added a new type of structure for the male genital organs to those already described by Looss. The simpler stage in the same type is furnished by the genus under consideration here. On the function of the male organs Looss says in a paper just received (01, p. 199) what is also implied in earlier contributions, namely, that the existence of an evaginable cirrus depends upon the presence of a muscular cirrus sac. There is in *Microphallus* absolutely no trace of a cirrus sac and there is also no modified terminal region of the duct which can be evaginated like that to which in other forms the name of cirrus is given. The species does possess, however, an ex-

trusible copulatory organ, which performs the same function as the cirrus and is consequently clearly analogous to it, though far from being homologous. I was mistaken in speaking of it in the earlier paper as "the morphological equivalent of the cirrus;" it is the physiological equivalent but is morphologically distinct. It will be an allowable inference that one may expect to find in *Levinseniella* a similar extrusion of the conical body in copulation. There are thus evidently two types of male copulatory organ in the distomes which depend in their function upon different mechanical principles. In the one the inversion of the duct in its terminal portion is brought about by the muscular cirrus sac in the manner described with care by Looss (94). This is evidently the more common among the distomes and in the interest of a precise nomenclature the name cirrus should be reserved for the eversible portion of the duct of this type of organ as Looss has already insisted. In the other type a permanent muscular (?) projection into the genital cirrus is developed about the terminal region of the duct; in copulation it is protruded and not everted. To this structure I have given the name copulatory papilla and Jägerskiöld has called it a conical body. Evidently both terms are individual in application, but the proposal of a definite name may well be deferred until the morphological value of the organ is better known and its occurrence among the distomes has been more fully investigated. Thus far it is known to occur only in the two forms discussed above. It should certainly not be designated a cirrus.

In one respect the description of Jägerskiöld is open to criticism. In the title he speaks of *Levinseniella* as a distome possessing a genital sucker, and while in the description he says that one can not well speak of a genuine genital sucker, he nevertheless emphasizes "a certain similarity of this organ with the true genital suckers." In my opinion this is clearly a confusion of terms. Morphologically it is not the enlargement or noticeable modification of the genital cirrus or yet the accessory copulatory structures that make a genital sucker, but the specific formation of folds and the development of muscles in the walls of the sinus which give it the structure and function of an acetabulum, as Jägerskiöld has clearly shown in other papers. Speaking of this form he specifically mentions that so far as he could find "the radial musculature of the genital sinus is wanting." In *Microphallus* as already

noted none could be demonstrated and the inert condition of the genital sinus during copulation goes to prove its absence, or at least that it is less well developed than ordinarily the case. This being so it is, I believe, incorrect to speak of a genital sucker here in any sense or to attempt a close comparison of these species with those in which such suckers exist. These copulatory organs evidently belong to a distinct type.

In this connection another point of terminology may be discussed. In my earlier paper (Ward, 94) the name *metraterm* was introduced to designate the modified terminal region of the uterus which had been variously designated by previous authors. Thus the term *vagina* has been used by writers on trematodes with reference to two regions morphologically distinct, the one of which, Laurer's canal, has been interpreted as the homolog of the vagina in other forms, while the other, the region now under discussion, is undoubtedly, as I was able to show in conjunction with others, the present functional vagina. Braun (93) had stated very clearly the objections to the term *vagina* and had met them by introducing a term "*Scheidentheil des Uterus*," which is incapable of rendition into a foreign language, save by a paraphrase. The distinguished investigator last cited has seen fit to make use recently of the name *metraterm*, which has also found favor with numerous other students of trematode structure. But Looss (94, p. 553-5) has discussed its use at length and has rejected it as unnecessary while he "retains the expression *vagina* for the region functional as *vagina*," etc. If one takes exception to this it must be on the ground implied in my earlier paper; the introduction of morphological terms has been wide spread and in the interest of greater precision; that Looss in his argument is compelled to speak of a "morphological *vagina*" and of a "physiological *vagina*" is to my mind good evidence of the confusion possible and of the need of a term which shall be at once brief and specific. The designation *metraterm* is a morphological one, applicable like *cirrus* to a definite structure in this group, and capable of consistent use for this alone. I think that it adds to the precision attained and that it may continue to find favor thereby.*

The genus *Levinseniella* as founded by Stossich included

*Similar views are advanced by von Ofenheim in a paper just received (Zeitsch. f. Naturwiss., Bd. 73, p. 161).

four forms: *D. brachysomum* Creplin, *D. pygmaeum* Lev., *D. macrophallos* v. Linst. and *D. opacum* Ward. To these Looss added later *D. claviforme* Brds. and as noted above removed *D. opacum* Ward. The latter is a fish parasite, while all the others are found in the caecum or rectum of birds. At the same time its structure differs considerably, being heavier, thicker and without spines, whereas the others are thin, delicate and covered with spines. The alimentary canal and suckers are more rudimentary, while one and all the reproductive glands are proportionally larger and heavier in *D. opacum*. In *Levinseniella* the uterus is smaller and the eggs less numerous, while the position of the reproductive organs varies somewhat. This is most noticeable in the separation of seminal vesicle and sexual pore by a long coiled ductus ejaculatorius in *Levinseniella*, while they are in close juxtaposition in *D. opacum*, and also in the structure of the copulatory organs. These facts may be brought together in the following description of the genus:

MICROPHALLUS n. g.

Small forms with flattened mobile anterior and thick immovable posterior region. Skin thick, tough, without spines. Prepharynx present, pharynx small, oesophagus long, crura very short, not even reaching end of anterior region. Excretory bladder large, V-shaped, reaching to the posterior limit of the testes. Ovary dextral, spherical, alongside of acetabulum. Seminal vesicle slightly pyriform, sinistral, composed of a single sac extending anteriorly from genital pore which is located on the left just behind the center of the acetabulum. All other reproductive organs behind ventral sucker entirely concealed by heavy coils of uterus filled with eggs. Testes two, directly behind ovary and vesicle. Vitellaria lobed, massive, lateral behind testes; vitelline ducts unite between testes to small vitelline reservoir below shell gland. Laurer's canal present but no receptaculum. Neither cirrus-sac nor cirrus present, but small conical copulatory organ developed about terminus of short ductus ejaculatorius. Eggs, abundant, 0.03 to 0.04 by 0.015 to 0.02 mm. Adult, parasitic in alimentary canal of fishes. Type and only species, *M. opacus* Ward 1894.

By virtue of the peculiar development which is shown by the copulatory organs this and *Levinseniella* may be grouped

into a new subfamily, Microphallinae, characterized particularly by the presence in the genital sinus of a muscular copulatory organ in which the ductus ejaculatorius terminates. In other respects the subfamily comes near the Brachycoelinae and Pleurogenetinae of Looss to the former of which it shows the greater similarity.

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EXPLANATION OF PLATE

The figures are made from camera drawings except as stated below.

ABBREVIATIONS

<i>c. p.</i>	Copulatory papilla.	<i>pr.</i>	Prostate gland.
<i>d. e.</i>	Ductus ejaculatorius.	<i>s. v.</i>	Seminal vesicle.
<i>e. r.</i>	Excretory reservoir.	<i>t.</i>	Testis.
<i>g. p.</i>	Genital pore.	<i>v. d.</i>	Yolk duct.
<i>int.</i>	Crura intestini.	<i>v. gl.</i>	Yolk gland.
<i>m.</i>	Metraterm.	<i>v. r.</i>	Yolk reservoir.
<i>ov.</i>	Ovary.		

Plate XXVI

Fig. 1. Ventral view of a specimen preserved in corrosive sublimate, stained in Czokor's alum cochineal and mounted in balsam. The heavy coils of the uterus were omitted to avoid confusion; they fall within the area bounded by the dotted line. The outlines are held true, but otherwise the figure is slightly diagrammatic.

Fig. 2. Cross section of pair in copulation showing relations of organs.

Fig. 3. Section showing seminal vesicle, ductus ejaculatorius, prostate cells and copulatory papilla; the latter is cut a little obliquely.

Fig. 4. Median section through copulatory papilla and genital sinus.

Fig. 5. Transverse section through *Levinseniella pygmaea* var. *similis* at the level of the genital pore. (After Jägerskiöld, 00, p. 735, fig. 3.) Magnification about 250.

PLATE XXVI

